

Fixation Protocol

Overview: This protocol is used for the fixation of cells in a cell-laden 3D bioprinted construct.

Materials:

- Paraformaldehyde (PFA). **PFA must only be handled inside a fume hood.**
- 50 mM CaCl₂ (CELLINK Cross-linking solution is 100 mM CaCl₂)
- Hank's Balanced Salt Solution (HBSS)
- Cell-laden 3D bioprinted constructs

Protocol:

All handling and use of PFA must be done inside a fume hood with proper PPE.

1. Wash cell-laden 3D bioprinted constructs in HBSS 15 min at 37°C
2. Fix the constructs in 4% PFA supplemented with 50 mM CaCl₂, 1h at RT
3. Wash the constructs 2x5 min with HBSS at RT
4. Store at 4°C in HBSS supplemented with 50 mM CaCl₂.

Further Information:

This protocol is optimized based on CELLINK™ Bioink, and may need further optimization for other bioinks.